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THE MONTANA STATE BOARD
OF ENTOMOLOGY

SECOND
BIENNIAL REPORT

1915-1916

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MONTANA STATE BOARD OF ENTOMOLOGY

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LETTER OF TRANSMITTAL

Bozeman, Montana, December 15, 1916.

To His Excellency,
Governor Samuel V. Stewart,
Helena, Montana.

My Dear Sir:

On behalf of the Montana State Board of Entomology I have the honor to transmit to you herewith the Second Biennial Report.

During the two-year period now closing there has appeared a widespread outbreak of Rocky Mountain Spotted Fever in a mild form, similar to that found in Idaho, in the eastern part of Montana. Roughly speaking this new infected area occupies the whole southeast one-fourth of the State. Numerous cases of the fever have occurred and several deaths. The Spotted Fever tick, which occurs in western Montana, is also abundant in this infested region in the eastern part of the State, and here, as in the Bitter Root Valley, is the agent of man's infection.

We have given preliminary attention to the Spotted Fever problem in Eastern Montana. A report of this work is presented together with a report of the work in the Bitter Root Valley and certain work on the housefly.

Very respectfully,

R. A. COOLEY,
Secretary.



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SECOND BIENNIAL REPORT

— OF THE —

Montana State Board of Entomology

By R. A. Cooley.

The activities of the Montana State Board of Entomology during the past two years may be classified under three heads, as follows:

1. Investigation and control of the Rocky Mountain spotted fever tick in Montana.
2. Investigation and control of the housefly in relation to human disease in Montana.
3. Miscellaneous preliminary studies of parasitic insects.

Control of the Rocky Mountain Spotted Fever Tick in Montana

Rocky Mountain spotted fever is, comparatively speaking, a new and little known disease of human beings. It is confined to America where it occurs, so far as at present known, only in nine Rocky Mountain and Pacific Coast States, as follows: California, Colorado, Idaho, Montana, Nevada, Oregon, Utah, Washington and Wyoming. Its occurrence and distribution have not been extensively studied in all of these states but, because of the severity and the number of cases in some of them, special attention has been compelled. Spotted fever is undoubtedly of more importance in Montana, Idaho, and Wyoming, than in other states, and of these three, Montana is looked upon as being particularly the home of the disease because of the fact that in this State, in the western part, is a locality where spotted fever occurs in a particularly fatal form.

During the past two years covered by this report the fever-tick situation has changed materially. Up to 1915 interest in spotted fever centered, so far as this State is concerned, almost entirely in the Bitter Root Valley and other territory close by. A few cases of much milder form had been reported from Carbon County and, although no official mention had been made of it, there was some reason to

believe that the disease existed in a restricted locality in the northern part of Gallatin County.

In the spring of 1915 suddenly, and without warning, spotted fever in a mild form appeared in several of the large counties in eastern Montana. This was the outstanding feature of the year and there were 22 cases in new territory in eastern Montana. Two of the cases, one in Big Horn County, and one in Fallon County, were fatal.

Human Cases of Rocky Mountain Spotted Fever in Montana in the Years 1915 and 1916.

Counties—	1915	1916
Western Montana.		
Ravalli	3	5
Missoula	4	1
Granite	---	1
Eastern Montana.		
Custer	8	1
Dawson	6	4
Rosebud	5	1
Big Horn	1	---
Fallon	2	---
Musselshell	---	2
Yellowstone	---	1
Fergus	---	1
Phillips	---	1
Scattering.		
Carbon	3	1
Gallatin	2	---
Cascade	1*	---
Totals	35	19

* This case came in from Idaho and did not originate in Cascade County.

In the course of the investigations conducted in 1916 it developed that 1915 had been a particularly bad year for ticks in Montana. An assistant spent some time in eastern Montana in that year and the observations made as compared with those made in 1916, as well as numerous statements made by residents showed that ticks were much more abundant the previous year. This probably accounts for the large number of cases in that year and for the dropping off of the number in 1916. However, it should be borne in mind that the infection in eastern Montana is a new one and we cannot

yet tell just how extensive it is and there is some reason to fear that the number may rapidly increase during the next few years. It is already clearly evident that the disease is spreading. A detailed account of these studies is presented in an article in this report by Assistant Entomologist, Dr. R. R. Parker.

Progress in the Eradication of the Rocky Mountain Spotted Fever Tick in Western Montana.

In the work of the spotted fever tick in the Bitter Root Valley both investigational lines and practical control are being pursued. As fast as information is obtained it is put into practical use in the field.

We feel that good progress is being made but on account of the long drawn out life cycle of the tick and the overlapping of broods, considerable time is required. There are now fewer ticks than formerly and fewer cases of fever. In earlier years the cases of the disease were running from nine to twenty-eight. Our work under the State Board of Entomology began in the spring of 1913. Data regarding the number of cases in that year are lacking, but in 1914 there were eleven, in 1915, seven, and in 1916, six cases. These figures taken alone give only an inaccurate impression of the progress that has been made for the disease has been spreading up and down the valley during recent years and has extended beyond where the control work has been taken up. The work of control was first undertaken where the situation was the most distressing, which was in the vicinity of Florence, Stevensville, and Victor on the west side of the valley. Here the work has been pursued with energy from the first and here, where the majority of all the cases formerly came from, we are now having very few. At the same time cases are springing up in the far upper part of the valley and in the far lower part of the valley, which fact, together with the occurrence of some cases up Lo Lo creek, where the work of control is not yet fully under way, accounts for a considerable part of the number of cases now occurring. Where the work is well under way, then, distinct progress is shown.

It should be borne in mind that no two localities are just alike with respect to the conditions which favor control of the tick. In each new locality it is necessary to get new in-

formation and conduct the campaign in a different way. The conditions up the Lo Lo creek are unlike those around O'Brien creek, and both of these are very unlike those around Florence, Stevensville and Victor.

It is clearly apparent that if no work of control had been taken up we would now be haing, with the increased cases up and down the valley, some 14 to 18 cases at least per year. instead of some six or seven, and it therefore appears that during the past four years some 20 to 35 lives have been saved and perhaps more.

The saving of life is of course the principal item of benefit derived from the control program, but this is not all. There can be no doubt that the presence of spotted fever disease in a locality has in past experiences worked out to be a detriment. Residence in such localities has been considered to be less desirable and property values have suffered. Towns to which infested localities are tributary have also been hurt in a business way. It is already apparent that in the parts of the Bitter Root valley where the work of eradication has been under way, there is a new confidence in the land value and in the locality generally.

REGULATIONS OF THE MONTANA STATE BOARD OF ENTOMOLOGY.

The complete regulations of the State Board of Entomology, with respect to the control of the Rocky Mountain Spotted Fever Tick in western Montana, are as follows:

Section 1.

The following tick-control districts are described and declared:

(1) The Florence District. Bounded on the east by the main channel of the Bitter Root river; on the north by the boundary between the 2nd and 3rd tiers of sections northward from the dividing line of Missoula and Ravalli counties, and extending from the river westward to and into the mountains as far as human habitations do or may go, or as far as domestic animals may wander; on the west by an imaginary line running north and south along the mountains as far back from the valley as human habitations do or may occur, or as far as domestic animals do or may wander; and on the south by the boundary line between townships nine and ten north, known as the O'Hare lane, and extending from above described imaginary line on the west, eastward to the main channel of the Bitter Root river.

(2) The Stevensville District. Bounded on the east by the main channel of the Bitter Root river; on the north by the boundary line between townships nine and ten north, extending from the main channel of the Bitter Root river to and into the mountains as far as human habitations do or may go or domestic animals do or may wander; on the west by an imaginary line running north and south along the mountains as far back from the valley as human habitations do or may occur, and as far as domestic animals do or may wander; on the south by a certain stream of water commonly known as Big Creek, extending from the mountains to the Bitter Root river.

(3) The Victor District. Bounded on the east by the main channel of the Bitter Root river, on the north by a certain stream of water known as Big Creek, extending from the Bitter Root river to and into the mountains; on the west by an imaginary line running north and south along the mountains, as far back from the valley as human habitations do or may go, or domestic animals do or may wander; on the

south by the highway locally known as the Bourne lane and on the east and west extension of the same, extending from the main channel of the Bitter Root river and into the mountains.

(4) The Hamilton District. Bounded on the east by the main channel of the Bitter Root river; on the north by Bourne lane and on the east and west extension of the same; on the west by an imaginary line running north and south along the mountains, as far from the valley as human habitations do or may go and domestic animals do or may wander; on the south by a certain stream of water commonly known as Canyon Creek extending from the mountains to the Bitter Root river.

(5) The Gold Creek District. Bounded on the east by the main channel of the Bitter Root river; on the north by a certain stream of water commonly known as Canyon Creek, extending from the mountains to the Bitter Root river; on the west by an imaginary line running north and south along the mountains, as far from the valley as human habitations do or may go and domestic animals do or may wander; and on the south by Lost Horse Canyon and creek and east and west extensions of the same.

Section II.

At a convenient point in each tick-control district shall be constructed a dipping vat and yards, for use in dipping domestic animals, and all cows, horses, mules, asses and dogs shall be periodically dipped or otherwise freed and kept free of ticks, by spraying, carding or hand-picking, at such times and in such manner as the officers in charge of the work may prescribe.

Amendment to Section II. The rules and regulations of the Montana State Board of Entomology, respecting the dipping of domestic animals for the destruction of the Rocky Mountain Spotted Fever tick are hereby amended; and on and after May 20th, 1913, or until otherwise notified, dairy cows which are giving milk are exempt from dipping, provided the owners keep them free of ticks by hand-picking or spraying with an arsenic-pine-tar solution, containing .22 per cent of arsenic trioxide.

Section III.

Owners of domestic animals in tick-control districts shall be allowed to dip their animals in the State dipping vats

without charge, but the responsibility of delivering the animals for dipping and of passing them through the vats, or otherwise freeing them of ticks, as prescribed by the men in charge of the vats, is placed upon the owners.

Section IV.

A close quarantine of all domestic animals, including driving horses, mules and oxen, shall be placed upon animals and premises of all persons who refuse or fail to bring their animals to the vats for dipping.

Section V.

On and after April 1, 1914, domestic animals, including cows, horses, asses, mules, sheep, goats and hogs, may be removed from the tick-control districts between March first and July fifteen of each year only on written permits of the Secretary of the Board of Entomology or duly authorized representatives of the United States Bureau of Entomology or the United States Public Health Service.

Section VI.

On and after April 8th, 1916, domestic animals, including cows, horses, asses, mules, sheep, goats and hogs, shall be prohibited from entering any tick-control district as designated by the Montana State Board of Entomology, for grazing or feeding purposes between February first and July fifteenth of each year, unless accompanied by a permit issued by the Secretary of the State Board of Entomology or a duly authorized representative of the United States Bureau of Entomology or the United States Public Health Service.

"Helena, Montana, April 5, 1914.

"TO WHOM IT MAY CONCERN:

"Notice is hereby given that Doctor L. D. Fricks, Victor, of the U. S. Public Health Service and Mr. W. V. King, Florence, of the U. S. Bureau of Entomology, are authorized to issue permits for the removal of domestic animals from tick-control districts.

"By authority of the Montana State Board of Entomology.

"R. A. Cooley, Secretary."

The cooperative arrangement with the United States Public Health Service and the Bureau of Entomology of the United States Department of Agriculture, as mentioned in the First Biennial Report of the State Board of Entomology, has been continued during the past two years with Dr. L. D. Fricks representing the Public Health Service and Dr. W. V. King representing the Bureau of Entomology as formerly.

The reports of Dr. King and Dr. Fricks to the Board of Entomology are herewith presented.

We have been very much in need of information regarding the germ or organism which is the specific cause of spotted fever. The Public Health Service through Dr. Fricks has been making a study of the virus and his results as first published in the Public Health Reports is attached.

The Board of Entomology has also engaged the cooperation of Dr. S. B. Wolbach, Associate Professor of Pathology and Bacteriology, Harvard Medical School of Boston. The results of Dr. Wolbach's studies are also presented.

Dr. Frick's and Dr. Wolbach's studies are not yet completed, but both are continuing the investigation. It is expected that Dr. Wolbach will also be in Montana next spring for the purpose of carrying on his work on the virus.

REPORT ON THE INVESTIGATION AND CONTROL OF THE ROCKY MOUNTAIN SPOTTED FEVER TICK IN MONTANA DURING 1915-1916.

W. V. KING, Ph. D.

Bureau of Entomology,
U. S. Department of Agriculture.

Following the general plans of the work as outlined in a previous report to the Montana State Board of Entomology¹, experimental control operations against the spotted fever tick (*Dermacentor venustus*) have been continued during the spring seasons of 1915-1916. This work has been confined to the northern half of the Bitter Root Valley in western Montana and the area includes several typical endemic foci of infection. Our efforts have been directed toward the determination of a practical plan by which the tick might be eradicated or at least reduced to the point of safety. The peculiar habits and host relationship of the spotted fever tick, combined with local conditions has made the problem a particularly difficult one, and the extremely long duration of the various stages in its life cycle has, as was to be expected, greatly delayed the demonstration of results of the control work. During the 1916 season, however, a comparison of the abundance of ticks at present with the abundance under normal conditions has shown a distinct improvement and given encouragement to further efforts.

The control program is based upon the fact that the tick must feed upon warm-blooded animals in order to develop and reproduce. Domestic animals are the usual hosts of the adult tick and the small native rodents are hosts of the immature stages. In brief, the general plan of the work has been; first, to prevent the engorgement of adult ticks on domestic animals as the principal measure; and, second, to supplement this with important secondary measures, principally the destruction of small native animals an encouragement of the clearing and development of tick infested lands. In organizing for the work, the principal foci of infection as indicated by the occurrence of human cases were selected. These have been divided into convenient units, the

¹—King, W. V., 1st Bien. Rept. Mont. State Board of Ent., Helena, 1919, pp. 16-27.

size of each being determined by local conditions. Three such districts were in active operation during 1914-1915, but in 1916 the work was discontinued in one of them, owing to lack of funds as well as unusual natural obstacles encountered. Toward the latter part of 1916 preliminary steps were taken to inaugurate the control work in a new district in which infections have been occurring recently.

In the present report the discussion of methods and results will be limited to the operations in the two districts in which the work has been uninterrupted for the three seasons. These are known as the Florence and Stevensville districts. The area included lies in the main valley and consists of about 28,000 acres of farming and timber lands exclusive of the mountain sides where stock may be grazed in many places. Approximately a half of this area is cleared and under cultivation, the other half being for the most part cut-over timber land and used as pasturage for domestic animals. This cut-over timber land lying along the foot of the mountain constitutes the area of tick infestation, as it is here that conditions favorable for their development exist.

From a census made in 1914, the two districts contained 119 farms with a total of 1,569 horses and 1,784 cattle. In the control work, however, a good proportion of these are eliminated from consideration as they are pastured outside of the tick infested fields.

Starvation and Destruction of Adult Ticks.

In the measures directed against the adult tick, no single method has been found which is practical under all conditions. Periodic dipping of cattle and horses, supplemented with hand-picking of small herds, of dairy cattle and work horses, and the removal of the stock from the tick infested fields have been employed according to the circumstances. The difficulties of the dipping program due to the necessity of short intervals between dippings, to the local conditions of the range and the ultimate expense have operated against the success of this method. These difficulties are being met by elimination of domestic animals from the tick country in the spring.

The active period of the adult tick lasts from the opening of spring, usually in March, until some time in June. The pastures and range are not suitable for use until about the first

of May and prior to this time the stock are or may be kept on the tick-free hay fields and meadows. The fact that the period is so short during which the use of the pastures is lost if the stock are kept out of them is favorable to the starvation method of tick control and in 1916 most of the stock owners were able, with our assistance, to secure other pasturage during the tick season. Some of the owners were able to provide tick-free fields large enough to accommodate their herds for this period and where possible this is undoubtedly the best plan. One owner found it advisable to reduce the number of his animals for this purpose. In one locality a district herd was organized and herding expenses paid jointly by the owners and the Bureau. This herd, consisting of about two hundred animals, was kept outside the control district and eliminated most of the animals which could not otherwise have been provided for. In another district a tick-free pasture near the river was secured to accommodate several small herds which had been very troublesome in previous seasons.

At a central point in each of four districts a dipping vat of about twenty-five hundred gallons capacity is located. One of these is of concrete and the other three of galvanized iron, the latter being preferred because of the ease with which they are kept in repair. If desired, they may be also moved. The first of the season, usually in March, these are filled with an arsenical dipping solution. If many animals are dipped during the season a second filling is necessary, usually in May. Sodium arsenite and a weak kerosene emulsion are now used in the dipping formula to replace the white arsenic-sodium-carbonate-pine tar formula in general use in the Texas fever tick work.

The method of preparation of the dipping solution is as follows: Sodium arsenite containing about 70% arsenic is used and the solutions are prepared so as to contain between .195 and .200% arsenic in the arsenious form. The capacity of the vat is computed by the prismoidal formula¹ and the amount of sodium arsenite needed to give the desired strength is figured from the actual percentage of arsenic in the sample, allowance being made for any variations apt to occur in each vat as determined by previous experience in mixing solutions in the different vats. The proper amount of

¹—See U. S. Dept. Agri., Farmers' Bull., No. 498, p. 36.

sodium arsenite having been determined and weighed out, is dissolved by heating in about 50 gallons of water. Arsenic in this form is readily soluble and is usually dissolved by the time the water has come to the boiling point. The solution is then added to half a vat full of water. Ten gallons of kerosene are emulsified, mixed with the solution in the vat and the vat then filled to the proper depth with water.

A considerable improvement in the preparation of the kerosene emulsion has been made as a result of tests made for us by the Chemistry Department of the Montana State Experiment Station. The details of the method are as follows: **Potassium** soap (green soap) is dissolved in warm water in the proportion of 12 pounds of soap to 2 gallons of water. This is allowed to cool and the emulsion is made in a cold solution at the rate of 3 gallons of kerosene to 2 quarts of soap solution. The emulsion is made in a hand spray pump, one in which the nozzle can be turned back into the tank. The soap is placed in the tank and pumped back until it begins to foam. The kerosene is then added, slowly at first, with forceful spraying of the mixture back upon itself. When the emulsion is complete, as evidenced by a thick, creamy consistency, it is diluted with water by mixing in a separate tank. The entire ten gallons of kerosene are emulsified in this way and the final mixture added to the arsenic solution in the vat.

After thorough mixing of the ingredients of the vat, an eight-ounce sample is taken and an analysis made to determine the percentage of arsenic present as arsenious oxide and total arsenic. (All of the analysis have been made by the Chemical Department of the Montana State Experiment Station, to whom I am greatly indebted.) No dipping is done until the analysis has shown that the proper percentage of arsenic is present. The strength of the arsenic is apt to be raised by evaporation or lowered by dilution with rain or oxidation of the arsenious oxide, so that frequent analyses are made and the strength corrected as necessary. It is sometimes advisable also to add more kerosene emulsion. The amount of kerosene present is too small to be of any especial value as a tick killing agent and is employed largely to give added penetration to the arsenic solution. Any kerosene which has not been completely emulsified and which may separate from the solution should be removed by skimming as it

floats to the surface. Free kerosene has been found to be injurious to the animals.

The tests under way to determine the exact effect of the arsenic solution upon this species of tick have not been completed.

Based upon the engorgement period of adult female ticks, the interval between dippings has been set for ten days. To be absolutely effective the interval should be even less, but since a ten-day period has been difficult to enforce it has seemed impractical to attempt to reduce the interval except in experimental work. The dippings are supplemented with hand picking.

The following numbers of animals were dipped and examined in the Stevensville and Florence districts in 1915 and 1916.

Year	No. Dipped			No. Examined		
	Horses	Cattle	Period Days	Horses	Cattle	Period Days
1915	142	1266	73	586	1267	109
1916	87	664	51	188	836	88

The reduction in number dipped and examined in 1916 is due largely to the removal of stock from the tick infested fields this year.

Destruction of Columbian Ground Squirrels.

Since it appeared unlikely that the breeding of adult ticks could be completely stopped, it was decided to reduce the development of the immature stages as much as possible by the destruction of their hosts, the small native rodents. The Columbian ground squirrel (*Citellus columbianus*) is the most important host of the larval and nymphal stages and is also an injurious pest of crops. In 1914 preliminary tests with poison grain were made and in 1915 and 1916 nearly all of the tick infested area in the two districts were given a treatment with the poison.

The poisoned grain formula which gave the best results is:

Hulled oats (groats)	16 quarts
Water	2½ pints
Salt	1 pint
Saccharine	1 teaspoonful
Starch	1 pint
Strychnine alkaloid (powdered)	2 ounces

The directions for preparation are as follows: Mix the saccharine and salt and dissolve in the two and a half pints of warm water. Add one pint of starch and heat (but not cook), with constant stirring until it becomes quite thick. Remove from stove and add the strychnine, which must be thoroughly mixed with the starch solution with an egg beater. Pour the mixture, now about the consistency of stiff whipped cream, over the 16 quarts of hulled oats contained in a tub and give thorough mixing by rubbing between the hands. The alkaloid of strychnine is insoluble in water and by this formula the grain is simply given a poisonous coating.

This was freshly prepared at the beginning of the season and its distribution begun as soon as the squirrels emerged from hibernation, since this is the most effective period of the year. In order to have the area covered as quickly as possible after the squirrels appear, it was divided into five parts and a crew of three men placed in each. In distributing the poison, a teaspoonful of the poisoned grain was placed at the opening of each squirrel burrow and records were kept of the number of baits placed and the amount of land treated. These operations were carried out each year during the first half of April.

The following tabulation is taken from records of the squirrel poisoning work:

Year	Total Acreage	Total No. Baits	Total No. Man-Days	Total cost of labor and poison
1915	9535	83,559	142.5	633.45
1916	8210	58,121	128.5	552.30
		Average baits per acre	Average acreage per man per day	Cost per acre
1915		8.7	66.9	.065
1916		7.1	63.9	.067

Effect of Poison. It is very difficult to approximate the proportion of squirrels killed by the poison. Under certain conditions, as in the vicinity of fields of young grain the squirrels do not take the poison well. In other places, and for the most part throughout the area treated, the poison was well taken and individual colonies were noted in which nearly all of the squirrels had been killed by the poison. Our

own observations and an inquiry among the residents familiar with the area treated led to the conclusion that at least half of the squirrels had been killed by the 1915 poisoning and a further reduction of about 50 per cent was roughly estimated to be the result of the treatment in 1916.

Results of Control Work.

In estimating the effect of the control measures in the reduction of ticks, several methods are employed. Unfortunately all of these are open to more or less criticism. The most promising method is one in which an index of the seasonal rate of infestation of ground squirrels is obtained and used for comparison. A series of these rodents from the tick infested area are killed (by shooting) and immediately placed in white muslin bags. The ends of the bags are tied to prevent escape of the ticks and the examination is made several hours later. By this time the larval and nymphal ticks have detached and are found crawling on the bag or in the hair of the dead animal.

In the following table is shown the infestation rate for three seasons:

Nymphal Infestation.

Year	No. Squirrels	No. Nymphs	No. Nymphs per squirrel	Reduction from	
				Normal	1915
Normal	181	863	4.76		
1915	333	232	0.69	85.6%	
1916	376	159	0.42	91.3%	39.3%

Larval Infestation.

Year	No. Larvae	No. Larvae per squirrel		
Normal	59	735	12.4	
1915	269	312	1.16	90.7%
1916	268	25	0.093	99.7% 92%

The "normal" is based on a series of squirrels examined in 1910. The most interesting comparison is found in the average nymphal infestation in 1915 and '16, as exactly the same area was covered in each season. The 1910 records were made in a portion of this area. The larvae commence

to appear on the squirrels the latter part of June and the average infestation is taken from the squirrels examined after the first larvae are found. The examination of squirrels is of further value in showing the exact points in which development of ticks is occurring.

The average infestation of domestic animals with adult ticks and the proportion of engorged ticks present is also employed as a method of estimating results, but is not of great value. Such data as are at hand are shown below. The numbers of engorged females per animals (per day) as given in the last column, are used for comparison.

Infestation of Cattle and Horses.

Year	Total No. Animals	No. Ticks	No. Eng'd Females	No. ticks per animal	No. Eng'd fem per animal
Normal	235	289	8	1.23	.034
1914	1720	622	11	.36	.0063
1915	1793 (a)	728	18	.4	.01
1916	1024	1648	9	1.6	.0087

The normal records were made in 1914 in a nearby locality in which conditions were similar to those in the control district. The data for the different years are not strictly comparable, since it is not possible to examine animals under the same conditions and with the same frequency each season. In 1916 it appears that the average number of ticks per animal for the whole district is greatly increased. As a matter of fact, this is due to the fact that a few animals in a small but heavily infested portion of the district were frequently hand-picked instead of dipped, as in previous years.

A third method of estimating results is the collection of unfed adults from the infested fields by dragging a piece of white flannel cloth about over the grass and brush where the ticks are awaiting hosts. Several factors such as wind, temperature, and time of season operate against the effectiveness of this method and the results are hardly comparative.

In 1915 and '16 records were kept of the numbers of ticks picked up by the squirrel poisoners. These figures are more or less comparable, as the number of men, area covered and time of year were practically the same.

(a)—The number of ticks not counted on 60 other animals examined.

Ticks Collected by Squirrel Poisoners

Year	Period	Number man day	Total ticks	Ticks per Man-day	Mean Temp.
1915	April 6 to April 17.	132	758	5.74	53°F.
1916	April 8 to April 17.	126	372	2.95	45°F.

It was indicated by this comparison that in 1916 the density of ticks had been reduced 48 per cent from the previous year. This was as great, in fact a slightly greater reduction than was to be expected from the 1915 record of the status of nymphal infestation of ground squirrels.

In interpreting the results, the complicated life history of the tick must be taken into consideration. Our studies have shown that under western Montana conditions this species has a two- and three-year life cycle. A reduction of the number of ticks engorged in any spring would result in an equal reduction of the larval ticks fed in the summer and of the nymphal ticks feeding the second spring, but would effect only a half of the active adults present during the third and fourth season, since the active adults are made up of two separate broods probably in nearly equal numbers. For example, a reduction of 85 per cent of engorged females in both 1914 and '15 should give a reduction of 42 per cent of active adults in 1916 and an 85 per cent reduction in 1917. The destruction of rodents is considered to effect the abundance of ticks in direct ratio to the percentage of rodents killed. The nymphal infestation of ground squirrels in 1915, combined with the estimated reduction of these animals, indicated an approximate reduction of 92 per cent of that brood of adults and a proportionate (46%) reduction of the total adult supply for 1916. Interpreting the nymphal index for both 1915 and '16 in the same way, a 94 per cent reduction of active adults is indicated for 1917.

Field Experiments.

In the way of experimental results, the most definite information has been obtained from observations made in a field of 320 acres which is being freed of ticks by the starva-

tion method. The field was known to be well infested with ticks in 1910 and '11. In 1914 it was fenced and has been kept entirely free of domestic animals for three seasons. About half of the field was cleared of timber and brush and 40 acres were put under cultivation. In addition to the elimination of domestic animals, squirrel poison was distributed in the field in 1915 and '16 with an estimated reduction of 60 to 75 per cent of these animals.

The following data have been recorded on the comparative infestation of this field, the results being attributed to the elimination of domestic animals and the destruction of squirrels.

Examination of Ground Squirrels in Experimental Field.

Year	No. examine.	Nymphs		Larvae	
		Total	Average	Total	Average
1910	10	59	5.9		
	6			43	7.1
1915	14	2	0.14	---	---
1916	10	---	---	---	---

The average rate of nymphal infestation for this vicinity in 1910 was 4.8 and for larvae. 12.4. It is therefore evident, although the number of squirrels examined is small, that the development of ticks is greatly reduced. The owner of the field, who is very familiar with the conditions there, stated that the reduction in adult ticks in 1916 was very marked. By another season it is expected that the full effect of the starvation method will be in evidence in the abundance of adults.

Sheep Experiments. Two experiments with sheep are under way to determine their status as hosts of the spotted fever tick. These were undertaken on the theory advanced by Fricks¹ that sheep might prove to be unsuitable hosts of the ticks because of the wool and lanolin oil secreted in the wool. One experiment is with a band of one thousand sheep on an area of about twelve hundred acres. No results have as yet been obtained from this experiment. In the second one a ten-acre field in an area of high tick infestation has been fenced and a few sheep were kept here in 1915 and '16.

¹—Fricks, L. D., U. S. Pub. Health Rep., 1913, pp. 1647-1653.

Frequent examinations were made and notes taken on the number of ticks present, state of engorgement and number killed by the lanolin in the wool or other factors. These records are given below:

Big Creek Sheep Experiment.

Year	Period	Sheep in Field		No. Examinations	Examinations		
		No. Sheep	Sheep-Days		Live Ticks	Dead Ticks	No. Fully Engorged live females
1915	 April 7	6	314	20	228	130	11
	to						
	June 16.	3					
1916	 May 3	6	300	24	371	141	8
	to						
	June 22.	6					
					1915	1916	
Average number live ticks per sheep.....					11.4	15.4	
Number engorged ticks per sheep per day....					.55	.33	
Number sheep per acre					.44	.6	
Period—days					70.	50.	
Number engorged ticks per acre for season					16.9	9.9	

(on basis of above averages.)

I have no available records to show the comparative development of ticks on horses and cattle under similar conditions. A series of 75 of these animals examined late in the season in 1914 showed an average development of one engorged tick per acre for the season. The locality was one of average infestation but much lower than in the experimental sheep field. From this fact and from general observations extending over several years, it is concluded that under such conditions the number of ticks which developed on the sheep were more than sufficient to maintain a normal supply.

Cooperation.

The experimental control operations are under the general supervision of Dr. W. D. Hunter and in close cooperation with the Montana State Board of Entomology. Professor R. A. Cooley, secretary of the Board, has been actively interested in the furtherance of the work and the Board has given financial assistance in the purchase of material for the squirrel poison. I desire to acknowledge further the helpful cooperation of the Montana State Board of Health, the U. S. Public Health Service, and the Bitter Root National Forest Service.

REVIEW OF ROCKY MOUNTAIN SPOTTED FEVER ERADICATIVE WORK CONDUCTED BY THE UNITED STATES PUBLIC HEALTH SERVICE IN THE BITTER ROOT VALLEY, MONTANA, 1915- 1916.

By SURGEON L. D. FRICKS.

The work of eliminating Rocky Mountain spotted fever from the Bitter Root Valley, Montana, as conducted by the United States Public Health Service has been continued during the past two years along the same lines as those indicated to the State authorities in my last report made at the close of the season of 1914. As a matter of fact there has been no alteration of nor retraction from the principles of spotted fever elimination as outlined by representatives of the Public Health Service soon after the work was begun by them in 1911. Some of these principles have been amplified however, as different investigations have thrown more light upon the subject of tick destruction.

These principles are as follows:

(1) The majority of the ticks which transmit spotted fever in the Bitter Root region breed on the domestic animals, horses and cattle, as they graze over the foot hills during the spring months.

(2) These uncultivated foot hills on the west side of the valley are now suitable only for grazing purposes and if possible should be so used.

(3) Because of practical difficulties which are unsurmountable, the dipping of domestic animals alone will not eradicate Rocky Mountain spotted fever from the Bitter Root Valley, and therefore the number of these animals, horses and cattle, should be reduced in every practicable way.

(4) The destruction of small rodents in the infected districts is highly desirable and the necessary expenditures justifiable.

All of the eradivative work which has been conducted by the Public Health Service in the valley has been based on these principles and may be briefly reviewed for the year 1915 and 1916 as follows:

Dipping of Domestic Animals.

The dipping of horses and cattle in the infected districts from Big Creek to Charlos Heights, a distance of thirty miles, was carried on during the tick seasons of the two years according to the regulations prescribed by the State Board of Entomology in so far as existing conditions would permit.

One of the greatest difficulties encountered in dipping in the Bitter Root Valley is the weather, which frequently makes it impossible to carry out the dipping of domestic animals at the necessary intervals during the tick season.

The season of 1916 was even worse than usual in this respect. Spring began early and the ticks were beginning to feed by the middle of March, but the cold weather dragged on from the first of April until mid summer. The latest record of its being too cold to dip during the year was on May 25th.

The annual outlay for the operation of the three dipping vats including chemicals and salaries of two vat tenders, but exclusive of the salary of a competent supervisor is in excess of \$1,000.

Results secured in tick eradication by the dipping of domestic animals which would warrant such an expenditure from year to year for the few hundred head of live stock involved have not been, and under the conditions which prevail in the Bitter Root Valley, cannot be obtained.

For this reason continuous effort has been made by the representative of the Public Health Service toward decreasing in every way by legislation, regulation and substitution, the number of horses and cattle allowed to graze on the west side of the valley.

Following several similar verbal requests made to the State Board of Entomology, letters were addressed to the Supervisor of the Bitter Root National Forest and the State Board of Entomology at the beginning of the 1915 season requesting that steps be taken by them toward the immediate reduction of all horse and cattle grazing and finally the complete abolishment of the grazing of horses and cattle for speculative purposes on the west side of the Bitter Root Valley.

In a reply received from the Supervisor of the Bitter Root National Forest he stated that in compliance with the request all permits for horses and cattle grazing on the

Forest reserve would be cancelled as soon as possible and no more would be issued.

Upon request and as an aid to the handling and dipping of domestic animals, a regulation was promulgated early in 1916 by the State Board of Entomology prohibiting the bringing into the infected districts for grazing purposes of domestic animals from February 1st to July 15th unless accompanied by a permit. This was a very necessary regulation for keeping account of the movement of domestic animals to and from the infected districts during the tick season; but it was not expected that such a regulation would reduce the number of domestic animals owned and grazed within the infected territory.

For this reason the request to the State Board of Entomology for additional legislation was repeated during the season of 1916 and it is to be hoped that an attempt will be made by the board to secure proper legislation this winter for restricting, so far as practicable the grazing of horses and cattle in the spotted fever districts of Southwestern Montana.

Sheep Substitution.

Because of the inadequacy of the dipping process as applied to horses and cattle in the Bitter Root Valley for the destruction of spotted fever ticks and for the several economic reasons enumerated in previous reports, experiments have been conducted during the past four years for the purpose of determining whether sheep could be satisfactorily substituted for the greater part of the horses and cattle now being grazed on the west side of the valley.

It has never been claimed that spotted fever ticks could not possibly breed on sheep under any and all conditions, nor has the turning loose of large numbers of sheep to roam at large among the west side foot hills been advocated by the Government Health officials; but it has been determined that sheep can be profitably grazed over this district in such ways as to decrease the tick infection.

During the past season six small bands were kept under observation and the results obtained in tick destruction were altogether good.

It has been necessary to inform the ranchers of the Bitter Root Valley of the progress of these sheep experiments

from time to time through the lay press, and they have been informed that the experiments are open to investigation or criticism from any interested party.

Small Animal Destruction.

During the past two seasons the principal methods of small animal destruction employed in the valley have been:

- (1) Poisoning.
- (2) Shooting.
- (3) Trapping.

All of these methods of destruction are justifiable under the existing Rocky Mountain spotted fever situation in the valley.

Poisoning. One thousand pounds of groats poisoned with strychnine furnished by the State of Montana were distributed over an area of 100 square miles, fifty by two, during 1915, and 1500 pounds during 1916.

It was found that the ground-squirrels (*Citellus columbianus*) would take this poison readily as soon as they are out in the spring and again during July. The outlay for poisoning, including cost of poisoned grain and salaries of ten men employed for from ten to fifteen days was approximately five hundred dollars each season.

It is not expected that the expenditure for labor will be continued indefinitely, but that after a few years the poisoned grain will be furnished to the ranchers and be distributed entirely by them. Figured on this basis the outlay should not amount to more than one hundred dollars per season.

Shooting. One hundred thousand rounds of small caliber rifle cartridges were used in shooting small rodents during the two years. Slightly more than half of this number was distributed to the west side farmers and used by them. The cost of the cartridges was small (twenty-three dollars per ten thousand) and the results worth the expenditure.

Trapping. In the Victor district where intensive rodent destruction was carried on a small force of trappers was employed both years. The method is expensive and only warranted here for experiment purposes. But the securing of a small number of traps, and their constant use by the west side ranchers has been strongly recommended.

ROCKY MOUNTAIN SPOTTED FEVER.

A Report of Laboratory Investigations of the Virus.¹

By L. D. Fricks, Surgeon, United States Public Health Service

During 14 years of investigation by different workers the following facts bearing upon the nature of the virus of Rocky Mountain Spotted Fever have been determined:

Man, rhesus monkeys, and at least six varieties of small wild rodents found in the Rocky Mountain region are susceptible to infection, while the larger domestic animals are generally immune. Of the laboratory animals, guinea pigs and white rats (*Mus norvegicus albinus*) are highly susceptible, while white mice (*Mus musculus albinus*) are apparently immune.

The virus is transmitted to susceptible animals, including man, by the bite of infective wood ticks (*Dermacentor*), recovery being followed by complete immunity. No other biting arachnid or other insect has been found capable of transmitting the virus. The transmission is not mechanical, since a tick once infected remains so, the virus multiplies in the tick and the female tick transmits the virus to her progeny. The virus may be propagated indefinitely in guinea pigs without loss of virulence by weekly blood inoculations, but dies within a few days outside the animal body. It will not pass through an ordinary Berkefeld filter under moderate pressure, and many attempts to cultivate it aerobically in the usual laboratory media have failed.

Wilson and Chowning, in 1902, described a piroplasm in the red blood cells of Rocky Mountain spotted fever cases seen in fresh blood smears both stained and unstained, but subsequent workers have failed to confirm their findings.

Ricketts reported diplococcoid bodies occasionally seen in fresh blood smears stained with Giemsa stain and many small bacilli found in infected tick eggs. He appears to have considered these as different forms of a specific microorganism, but afterwards found similar bacilli in noninfected tick eggs. Ricketts reported the agglutination of this bacilli found in tick eggs by immune guinea pig serum, in dilutions of 1 to 320, but was unable to cultivate the organism.

¹ Reprint from the Public Reports, Vol. 31, No. 9, March 3, 1916. pp. 516-521

Recent Investigations of the Virus.

In connection with the field campaign conducted by the Public Health Service for the purpose of ascertaining the measures best adapted to the eradication of Rocky Mountain spotted fever from a community, and for determining the present areas of infection in the Rocky Mountain region, laboratory investigations of the virus have been carried on both in the field laboratory at Victor, Mont., and at the Hygienic Laboratory, Washington. It is believed that the findings are of sufficient interest to warrant a preliminary report thereon at the present time.

All attempts made to cultivate the virus on many different media aerobically have failed, despite the fact that the virus circulates freely in the blood stream, 0.1 c.c. of blood frequently being sufficient to infect a guinea pig.

Attempts were made two years ago to grow the virus anaerobically by mixing infected guinea pig blood with freshly melted and properly cooled glucose agar and glucose ascitic agar, in different dilutions, with and without the addition of normal guinea pig kidney. No uniform results were obtained; occasionally, however, anaerobic diphtheroid bacilli were encountered, but inasmuch as they were all found nonpathogenic for guinea pigs they were abandoned.

Following the announcement of the discovery of the "*Bacillus typhi exanthematici*" by Plotz, and because of the close clinical resemblance between Rocky Mountain spotted fever and typhus fever, the different anaerobic bacilli, referred to above, which have been encountered since in cultures, have been studied more carefully.

Ten strains of these anaerobic bacilli have been isolated; some from dilute guinea pig serum plus normal guinea pig kidney planted with infected blood, some from glucose ascitic agar plus normal guinea pig kidney planted with infected blood, and one from freshly boiled 1 per cent glucose broth in fermentation tube in which 5 c.c. of infected guinea pig blood had been planted.

These bacilli have not been found with anything approaching the frequency with which Plotz was able to recover "*Bacillus typhi exanthematici*" from typhus cases, but in the writer's routine work only from 5 to 10 drops of infected blood were used for planting, after the chest wall has been opened, that amount being well above the minimum infective

dose for guinea pigs. Plotz, on the contrary, regularly used 2.c.c. or more of typhus blood, making the puncture through the skin.

All of the 10 strains referred to are strict anaerobes, growing equally well in deep stabs on freshly melted glucose agar and ordinary agar, and in fresh glucose broth in fermentation tubes.

These organisms are nonpathogenic to guinea pigs, are not agglutinated with immune guinea pig serum and do not show complement fixation with immune serum when used as antigen. The macroscopic method of agglutination was easily employed with cultures grown on fresh glucose broth in fermentation tubes. (Microscopically an apparent clumping of the bacilli is nearly always seen.)

These organisms, recovered from spotted fever guinea pigs, resemble very closely morphologically and culturally the two strains recovered from typhus guinea pigs by Hasseltine and Neill at the Hygienic Laboratory and the strain of *Bacilli typhi exanthematici* kindly furnished by Dr. Plotz.

Anaerobic Fluid Media Cultures.

In an endeavor to cultivate the Rocky Mountain spotted fever virus in fluid media under lessened oxygen pressures, the following technique was evolved at the field laboratory, Victor, Mont.:

Articles required:

Ten c.c. homeopathic vials, rubber stoppers to fit.

One-fourth inch glass tubing in 6-inch lengths.

One hand vacuum pump.

A constriction was drawn in the glass tubing, and a small hole, into which the tubing would fit snugly, was burnt in the rubber stopper. The stoppers with tube inserted and the vials were sterilized separately. After the vials had been filled with 8 c.c. of media and inoculated, the stoppers were driven in tightly and sealed with paraffine, and the glass tube attached to the hand pump. After five to 30 minutes' exhaustion the glass tube was sealed at the constriction previously made and the culture then placed in the incubator.

While there was no exact measure of the vacuum obtained, or of its duration, it was possible to inhibit completely the growth of aerobic organisms by this method when so desired.

The media used were human serum and guinea pig serum with normal salt solution in different dilutions (1 to 2 and 1 to 3) and ascitic fluid undiluted; a piece of fresh guinea pig kidney was added at the time of inoculation in practically all instances. The material employed in inoculating the media consisted of infected guinea pig blood and tissues, blood from human cases of spotted fever, and infected tick eggs.

The following is a brief summary of the results obtained: Forty-five series of vials were planted. In the beginning so much time was consumed in searching smears made from the cultures for microorganisms that it was decided to depend entirely upon animal inoculations, followed by immunity tests, in order to determine if possible the presence of the living virus in the cultures.

Ninety-seven guinea pigs were inoculated from these cultures and later tested for immunity by the injection of 0.5 c.c. of known spotted fever virus.

The cultures tested were from two days to one month old, the majority being less than two weeks old. All the guinea pigs injected with cultures less than two weeks old, when later given the immunity test, developed spotted fever.

Three guinea pigs out of ten inoculated with cultures 21 to 25 days old, either showed definite lesions of spotted fever or were immune to the spotted fever virus, as shown below:

Series No.	Culture Medium	Material Planted	Method	Animal Inoculations	Immunity Tests	Remarks
C1—June 15, 1915.	8 c.c. dilute human serum + normal g. p. kidney	5 drops seventh day g. p. blood.	Air exhausted, vial sealed & kept at 37°.	July 7, 5 drops injected into g. p.	G. p. immune to spotted fever virus injected June 30 and again on Aug. 6, 1915.	G. p. showed temperature above 40° C. June 17 to 22, 1915.
D2—June 17, 1915.	do.....	7 drops human spotted fever blood.	do.....	June 29, 5 drops injected into g. p. D2a, July 9, 5 drops injected into g. p. D2b.	G. p. D2a injected Aug. 28 developed spotted fever. G. p. D2b injected Aug. 13 and found immune.	G. p. D2b showed characteristic lesions of spotted fever following injection of July 9, 1915.
H3—July 15, 1915.	do.....	1 infected tick eggs crushed.	do.....	Aug. 10, 5 drops injected into g. p.	G. p. immune to spotted fever virus injected Aug. 28, 1915.	

Inasmuch as the Rocky Mountain spotted fever virus ordinarily dies within 24 to 48 hours when kept at a tem-

perature of 37°, as the dosage of the cultures injected was much less than the minimum infective dose of fresh virus, and as in the inoculation of several hundred guinea pigs no naturally immune guinea pig has been encountered, it seems reasonable to conclude that a multiplication of the virus occurred in the cultures C1, D2, and H3.

Centrifugation of the Virus.

Ricketts was unable to throw down the virus from guinea pig and monkey serum when diluted with equal parts of salt solution, even after prolonged centrifugation (six hours.) Centrifugation with greater dilutions of normal salt solution has been employed by the writer several times successfully for the purpose of freeing the virus from a coccus contamination. In this way, by injecting different layers of the centrifugal material, a layer was found which would produce spotted fever in the inoculated guinea pig. without carrying over the contaminating coccus.

By increasing the dilution to 1 part of serum to 8 or 10 of salt solution it was found that the spotted fever virus could be thrown down completely by four to six hours' centrifugation, as is shown in the following experiment:

After defibrinating 10 c.c. of spotted fever blood and centrifugating for 15 minutes, 1 c.c. of the serum was pipetted off and diluted with 10 parts of normal salt solution. This was then centrifuged for four and one-half hours at about 2,000 revolutions per minute. Ten c.c. of the supernatant fluid was carefully drawn off and injected into guinea pig S2. This pig showed no reaction whatever, and later developed spotted fever when inoculated with the virus; while guinea pig S1. inoculated with three drops of the sediment, developed spotted fever on the eighth day following inoculation and showed all the characteristic lesions of the disease as seen in the guinea pig. This experiment has been repeated many times with similar results.

Microscopical Examination of Spotted Fever Blood.

Stimulated by the fact that the virus of Rocky Mountain spotted fever does not pass through a Berkefeld filter (N) under a pressure of 1 atmosphere, various investigators of the disease have spent much time in searching fresh blood smears for the causative organism, but without agreement as to findings.

During the last three years the writer has examined many blood smears prepared and stained by all the well-known methods, from human cases, and from the known susceptible animals, particularly guinea pigs and white rats. Frequently there have been found in spotted fever blood, stained by the Giemsa method, extra corpuscular granules, singly and in pairs, staining bright red and highly refractile; also, similar bodies within or in close proximity to the erythrocytes. The intracellular bodies are usually surrounded by a pale halo. The presence of these granules was considered significant, but it has been impossible to differentiate them with certainty from the granules sometimes found in normal blood.

By dilution and centrifugation a method for concentrating and distinguishing these bodies appears to have been found, the best results having been obtained in the following manner: Ten c.c. of infected blood is withdrawn by heart puncture, defibrinated, and immediately centrifugated for 15 minutes. One c.c. of the surface serum is then pipetted off and diluted with 10 c.c. of normal salt solution in an ordinary centrifuge tube. One c.c. of the remaining serum, containing some of the upper layer of red cells, is treated in the same way. These fluids are then centrifuged for 6 hours, the supernatant fluid is carefully poured off, and smears are made from the drop of sediment remaining and stained over night with dilute Giemsa stain.

The serum smears show many bright red granular bodies, singly and in pairs, highly refractile, accompanied by larger light-blue bodies, and all surrounded by a pale-blue matrix, the whole mass being rather indistinct but not encountered in the controls. The red blood cells appear to take the stain normally, but in many of them are found round or slightly elongated red chromatin bodies partially surrounded by or in close approximation to a somewhat larger deep-blue staining body. Some of the chromatin bodies approach 1 μ in diameter, but the majority are smaller and in these the protoplasm is elongated, extending well beyond the chromatin body at both ends.

Some of the bodies are found clearly without the cells and in the largest of these the red chromatin body is centrally located and surrounded entirely by the deep-blue staining protoplasm, the whole being crescentic in shape.

This method of preparing and staining blood smears has been repeated many times with proper controls of normal guinea-pig blood and with blood from pigs sick with diseases other than spotted fever, with the result that the bodies above described have never been found except in spotted fever blood.

There appears to be some resemblance between these bodies found in spotted fever guinea pig blood and those described by Seidelin as having been found by him in yellow fever blood; and in view of the criticism of Seidelin's work made by Wenyon and Low, who claim to have found similar bodies in normal guinea pig blood, one naturally hesitates to draw any definite conclusions from the finding here reported.

From the fact that these bodies, on account of their morphological and tinctorial characteristics, may be regarded as probably of protozoan nature, and because they have thus far been found only in blood from animals infected with spotted fever, it is felt that the publication of their description at this time is justified, in order that other workers may be on the lookout for them, and that their relationship to Rocky Mountain spotted fever may be fully established.

The writer is indebted to Surg. A. M. Stimson and Asst. Surg. R. R. Spencer for assistance in carrying on the above-described investigations.

THE ETIOLOGY OF ROCKY MOUNTAIN SPOTTED FEVER.

By S. B. Wolbach, M .D.

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Boston, Massachusetts.

This study was begun in response to an invitation on the part of the State Board of Health and Board of Entomology of Montana to investigate Rocky Mountain spotted fever in Montana during the spring of 1916. In anticipation of that visit laboratory work was begun in January, 1916, in the bacteriological laboratory of the Harvard Medical school with the virus of the disease obtained from Surgeon L. D. Fricks. It was not possible to visit Montana because of illness, but with the exception of three months the investigation has continued to the time of writing. using laboratory animals, guinea-pigs and monkeys. The writer wishes to express his thanks to Surgeon L. D. Fricks for sending infected ticks and blood from infected guinea-pigs, to Doctor W. F. Cogswell, Secretary of the State Board of Health of Montana, and to Professor R. A. Cooley, State Entomologist of Montana for many courtesies and for the receipt of wild ticks—*Dermacentor andersoni*. The present report is essentially a summary of findings to date.

Pathology and Occurrence of the Parasite in Guinea-Pigs and Monkeys.

Rocky Mountain spotted fever as reproduced in monkeys and guinea-pigs has its characteristic lesions acute processes in the superficial blood vessels and blood vessels of the genitalia. The lesion is an acute endophlebitis and acute endarteritis, leading to proliferation of endothelium and thrombosis, often resulting in complete obliteration of blood vessels of the skin and testes. The lesions of the blood vessels are responsible for the rash, the oedema, hemorrhages, and the necroses. They correspond with the finding briefly rescribed by LeCount of Chicago, who worked with material obtained from Doctor Ricketts. It is unfortunate, however, that in the human no study was made of the peripheral blood vessels. The character of the lesion resulting in complete or partial

occlusion of small arteries and veins explains the character of the rash in the human, its long persistence and the occasional sloughing of the skin which has been observed particularly upon the genitalia.

In the vascular lesions in monkeys and guinea-pigs there has been found constantly in a large series of animals a minute parasite. This organism commonly occurs in the form of paired gramules or exceedingly short rods in pairs. It occurs in large numbers in many lesions. The organism is easiest demonstrable in sections of tissues which have been fixed in Zenker's fixative and stained with Giemsa's stain. They may also be demonstrated in sections fixed in Zenker's fixative and stained in Loeffler's alkaline methylene blue at 55°C. for 24 hours and differentiated in a one to one thousand solution of acetic acid in water. The organisms may also be demonstrated in smear preparations made by scraping affected tissues with a very sharp knife held vertically. Considerable patience is required to disintegrate the blood vessels by repeated scraping, because it is necessary to obtain isolated endothelial cells and smooth muscle fibres in order to obtain satisfactory preparations.

The greatest numbers of the organisms occur in the lesions of arteries and veins in the testicle and its appendages, in the cremasteric muscles and in the skin and subcutaneous tissues. One of the best regions for demonstrating the organisms in smears is the tissues of the paws (feet) after these become swollen, towards the end of the febrile period in guinea-pigs.

There is considerable variation in the size of the organism as found in the guinea-pigs.—a larger form which occurs as a lanceolate paired organism and a much smaller oval form which is found closely packed in smooth muscle fibres of the vessel walls and occasionally within cells in proliferated endothelial (epithelioid) cells lying in the adventitia of blood vessels. In thick film preparations of the blood from infected guinea-pigs and monkeys only one type of the organism may be found. This is the larger lanceolate rod in pairs. Ricketts undoubtedly saw these organisms and described them as having the form of "two somewhat lanceolate chromatin staining bodies separated by a slight amount of eosin staining substance." In the present study these organisms have not been seen in blood except in thick film preparations,

and therefore subjected to the action of distilled water. The technic of making thick film preparations is as follows: One or two large drops of blood are distributed on a microscope slide over an area 1 cm in diameter. After the film has dried it is de-hemoglobinized in distilled water; two or three changes are necessary and the preparation must be handled gently in order not to detach the film. When the preparation has become colorless it is dried in the air, fixed for fifteen minutes in absolute alcohol, dried and stained with Giemsa's stain. Subjected to the above treatment the organism occurs in pairs and consists of two deeply stained oval or lanceolate bodies colored deep red or purple and surrounded by a small amount of bluish stained material. It would appear as if this type of the organism were rich in chromatin material, if the coloration by Giemsa's method may be accepted as a criterion. The organisms in the blood are never very abundant, but they usually can be found in thick film preparations after a search of from five to fifteen minutes. They are most abundant in the first days of fever. In smear preparations from infected tissues, forms similar to those found in thick film preparations of blood are encountered, as well as much smaller oval and rod-shaped organisms which stain much less deeply and which take a bluish coloration with Giemsa's stain. Normal guinea-pigs and guinea-pigs infected with a strain of Typhus (Brill's disease) obtained from Doctor Peter Olitsky of the Mt. Sinai Hospital of New York were used as controls in studying the occurrence of the parasite in the blood stream; always with negative results. All attempts at cultivating this organism from guinea-pigs have failed, and many methods were employed, using media suitable for the cultivation of spirachætes, bacteria and protozoa. The organism has, however, been studied in infected ticks, and its presence is characteristic of and exclusively a feature of infective ticks.

Occurrence of the Parasite in Ticks.

Infected ticks were secured by allowing them to feed one or more times upon infected guinea-pigs. Their infectivity was subsequently proved by allowing them to feed upon normal guinea-pigs. Ticks which were proved to be non-infective by feeding once or twice upon normal guinea-pigs were used for controls. It was found that a single feeding often did not render a tick infective. Occasionally

two or even three feedings upon an infected guinea-pig were necessary to make a tick infective.

In testing for infectivity of the ticks, each one was confined in a close meshed wire gauze capsule fastened with adhesive plaster to the shaved abdomen of a guinea-pig. From two to five days were allowed for each feeding. The capsules served also to retain the feces passed by the ticks, the amount of which in each experiment offered an additional index of the amount of blood ingested. It was observed that the feces collect in the form of small balls which soon become hard and dry and do not soil the skin of the guinea-pig.

The procedure usually employed in examining the ticks, infested and control, was as follows: The dorsal surface was removed as carefully as possible under the dissecting microscope. From one-half of the tick, salivary gland, Malpighian tube, leg muscle, and a portion of the intestinal diverticulum were removed and smear preparations were made which were stained with Giemsa's stain, as well as preparations for the microscope with dark field illumination. The remainder of the organs were then removed in one mass from the ventral plate. The capitulum is left attached to the organs for the purpose of handling during fixation and imbedding processes. The tissues were fixed in Zenker's fixative, imbedded in paraffin and serial sections made, which were stained by Giemsa's method.

In ticks which were proved to be infective, parasite identical with those found in the tissues of guinea-pigs and monkeys were found. Both types of parasites occurred, the minute form predominating. In no instance were the parasites found in ticks which were proved to be non-infective.

The distribution of the parasite in the infected tick is wide and subject to some variation. They are found most abundantly in striped muscle, although occasionally they may occur in enormous numbers in the Malpighian tubes, both in the epithelial cells and in the lumina of the tubes. They are very numerous in the ganglion, or brain, in the main nerve trunks, in the salivary glands and walls of the salivary gland ducts. They are numerous in the smooth muscle fibres of the uterus and vagina. They are occasionally very numerous in the esophagus and are always to be found sparsely distributed throughout the intestinal tract. They have been seen in spermatozoa and ova.

It is worthy of note that there is no cellular reaction on the part of the tick to the presence of these parasites, even when they are present in enormous numbers. A similar absence of reaction to the presence of a parasite, *Spirocheta duttoni*, was observed by me in the tissues of the African tick, *Ornithodoros moubata* (Jour. Med. Research, Vol. XXX, o. 1.) The morphology of the parasite is identical with that found in guinea-pig and monkey tissues. It is possible now definitely to state that there are two principal morphological types—one, a lanceolate diplococcoid organism, which is considerably larger than the other type, a small, rather slender rod-shaped form. The lanceolate type is also found in the circulating blood and stains fairly deeply with the chromatin coloration, that is, reddish purple; at each end there is a small amount of pale blue staining material. The smaller, rod-like form stains bluish, or bluish purple, according to variations in the staining technic, and may contain granules, bi-polar or more numerous, as were described in preparations from guinea-pigs. It is obviously poor in chromatin staining material. To these types may be added a third, which, however, probably should be included with the rod form. It is a minute oval coccoid form which ranges in size down to forms just visible with the best optical equipment. They stain bluish with Giemsa's stain and have been found most abundantly in smear preparations of infected ticks. They are also to be seen in the sections in masses within cells, too compact to be resolved except at the periphery of the clumps. They occur also inside the nuclei of certain cells, namely the epithelial cells of the Malpighian tubules. Greatly distended nuclei are occasionally found filled with minute paired granules. Ruptured nuclear membranes filled with these masses have been found.

No new light has been obtained in regard to the exact nature of this organism. The arrangement in pairs, end to end, and in tissues, in chains of considerable length, supports the evidence that division occurs by transverse fission, as in bacteria. The lanceolate chromatin-rich form is the only type found with any constancy in the circulating blood of infected animals, and it is reasonable to suppose that this represents a more resistant stage of the organism, although no proof has been obtained on this point. The organism as observed in suspensions of crushed tissues of the tick by

dark field illumination is non-motile. It does not retain the stain by Gram's method.

Summary and Conclusions.

An organism having definite peculiar characteristics may be found in large numbers in the lesions characteristic of spotted fever in experimental animals, guinea-pigs and monkeys. These lesions are essentially proliferative in character. The cells which respond in largest numbers to the action of the organisms are endothelial cells. These accumulate in great numbers in the vessel walls and around the vessels. They may be seen in mitoses in various locations, in lymphatics and in blood vessels. While this organism has not been cultivated in artificial media it has been proved to multiply in ticks—*Dermacentor andersoni*—fed upon infected guinea-pigs. The organism in ticks is morphologically indetical with the organisms as found in the lesions of guinea-pigs and monkeys, although the minute chromatin-poor forms are much more abundant.

This parasite does not occur in the tissues of ticks proved to be non-infective.

The distribution of the parasite in the infected ticks indicates that the transmission occurs by way of the salivary gland secretion. Transmission by fecal contamination of the wound caused by the tick in feeding does not seem possible because of the character of the tick's feces.

At the present time it is impossible to classify this organism. Individually the organisms as they occur resemble bacteria. The very definite occurrence of two morphological types, one exceedingly minute, usually occurring in great masses and poor in chromatin, the other occurring in smaller numbers, rich in chromatin, larger in size, and having the peculiarity of being the only form that is found in the blood stream of infected animals, would indicate the existence of a more complicated life cycle than is common to bacteria. The peculiarities in the distribution of the organism in the tissues and the staining reaction are reminiscent of these characteristics in spirochaetes. It is quite possible that we are dealing with a wholly new type of micro-organism.

The facts presented in the above report have been adequately controlled in regard to the distinctive character of the lesions in experimental animals and in regard to the

specificity of the parasite described in experimental animals and in ticks. While it is highly desirable to confirm the presence of the parasite in human lesions, this evidence is not essential for the conclusion that the parasite described is the causative agent of Rocky Mountain spotted fever.

Description of Plates.

- Plate 1. Fig. 1. Arteriole of skin of guinea-pig showing parasites in the smooth muscle cells. Photomicrograph, 2000 diameters.
- Fig. 2. Small vein of testicle of guinea-pig showing parasites in the endothelium and smooth muscle fibres. Photomicrograph, 2000 diameters.
- Plate 2. Fig. 3. Smear preparation from salivary gland of infected tick; shows many parasites in the neighborhood of the nucleus, which has been partly disintegrated in the making of the preparation. Photomicrograph, 2000 diameters.
- Fig. 4. Paired lanceolate type of organism found in circulating blood. Thick film preparation. Monkey. Photomicrograph, 2000 diameters.
- Fig. 5. Paired lanceolate type of organism found in circulating blood. Thick film preparation. Guinea-pig. Photomicrograph, 2000 diameters.
- Fig. 6. Smear preparation from teased tissue of guinea-pig. Shows many organisms in endothelial cells. Photomicrograph, 2000 diameters.
- Plate 3. Fig. 7. Salivary gland of infected tick showing the parasites in a gland acinus of the second type. Camera lucida drawing from a section, 1500 diameters.
- Fig. 8. Portion of wall of intestinal diverticulum of an infected tick, cut tangentially. Shows scattered lanceolate forms throughout the muscle coat and one smooth muscle fibre packed with the smaller type of the organism. Camera lucida drawing from a section, 1500 diameters.

PLATE 1

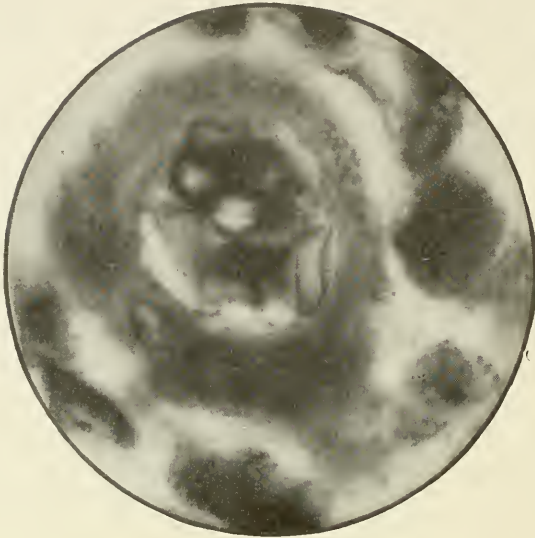


FIGURE 1

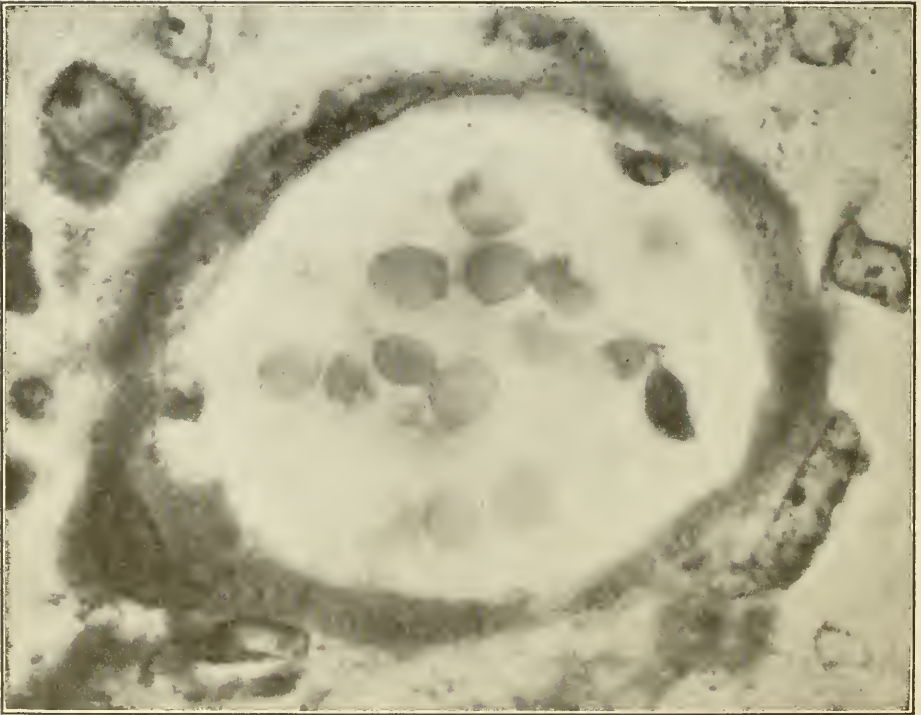


FIGURE 2

PLATE 2

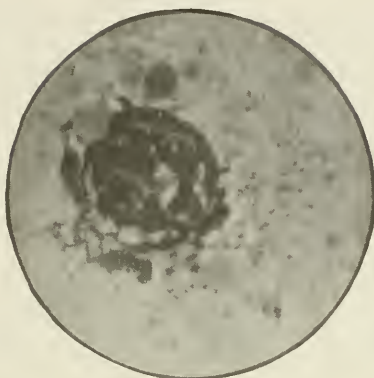


FIGURE 3



FIGURE 4



FIGURE 5



FIGURE 6

PLATE 3



FIGURE 7



FIGURE 8

**SOME FACTS OF IMPORTANCE CONCERNING THE
ROCKY MOUNTAIN SPOTTED FEVER TICK, (DER-
MACENTOR VENUSTUS BANKS,) IN
EASTERN MONTANA.**

By R. R. Parker, Ph. D., and R. W. Wells.

As the result of a week's preliminary survey of tick conditions in eastern Montana in 1915 it was found that the investigations previously carried on by the State Entomologist and the United States Bureau of Entomology in the Bitter Root Valley threw but little light on conditions in the former portion of the State and that control measures adapted to western Montana conditions were inapplicable. Therefore, during the season of 1916 a field station was established at Powderville on the Powder River in territory known to be infected, and studies were made of the bionomics of the tick and of economic conditions that would need to be considered in a control program. Actual investigations were carried on from March 27 till September 1. It should be borne in mind that the region studied was relatively very small in comparison with the extent of tick infested territory and that some of the results will be of general interest, some only of local import.

The results of the investigation may conveniently be discussed under the following headings: (1) topography, (2) character of vegetation, (3) species of ticks found, (4) tick abundance, (5) economic conditions bearing on control, (6) wild mammals as hosts of the spotted fever tick, (7) domestic animals as hosts of the spotted fever tick, (8) ticks on human beings, (9) life history notes, (10) longevity experiments, (11) means by which ticks are spread.

Topography.

The country in general may be said to consist of river and creek valleys and of hills, the hill country being by far the most extensive. In the section studied the valleys are comparatively narrow and consist of bottom land, that which the river has most recently left, and of bench land, which extends from the bottom land to the hills. The bench land is usually of greater extent. Creeks cutting across the valleys to the river wear deep gullies, many of which are dry during the summer except after heavy rainfalls.

Character of Vegetation.

Sage brush is the most common plant life. It grows most luxuriantly on bottom land and wherever on other types the soil is damp. It is abundant on bench land, much less common on the hills. Considerable stretches of bench land, however, may be barren except for sparse grassy growth.

Trees are scarce and likely to decrease further with the increase of settlers. Open stands of cottonwoods are found along the rivers and in some places occupy considerable portions of the bottom land, especially in bends of the river. In such places heavy undergrowth of brush is not infrequent. Cottonwoods are also sometimes found fringing creek beds. Shrubby growth occurs along some of the creeks but is not dense. Occasional hills and ridges occur that bear very open stands of scrubby pine.

Species of Ticks Found.

Several species of ticks were found on a variety of host animals, wild and domestic. The most abundant species was the wood tick, *Dermacentor venustus* Banks, the only agent known to transmit Rocky Mountain spotted fever in nature. Other species of ticks encountered were the rabbit tick, *Haemaphysalis leporis-palustris* Packard, the spinose ear tick, *Ornithodoros megnini* Duges, and species of *Ixodes*, the most abundant of which was *Ixodes kingi* Bishopp. It was suspected that at least one other species of wood tick, the eastern dog tick, *Dermacentor variabilis* Say, might be found. Though not met with in the vicinity of Powderville, several specimens taken from men's clothing at Kirby, in Big Horn County, were received by Professor Cooley. Specimens were also received by us from North Dakota, where it is reported as abundant and frequently found on man.

Tick Abundance.

Wood ticks have been known to the residents in southern Custer County for at least 25 years and probably have been there even longer. As far as can be judged on hearsay evidence, they have increased somewhat in abundance. One of the most interesting developments is the fact that there are occasional years of unusual tick abundance. The season of 1915 was of this character with the difference that the ticks were more numerous than at any previous time recalled by the ranchers. The last previous season of great

abundance was said to be during 1908. Between these seasons of great abundance were intervening years during which ticks were relatively scarce, though possibly gradually increasing in numbers. The factors which have determined this great variation in numbers are important to determine, but are obscure at present; climatic conditions, availability of hosts for the immature stages of the tick and for the engorgement of females must all be considered and correlated. The above statement of occasional tick abundance should be qualified to this extent, that though it seems to hold true for the largest portion of the section studied, there are, nevertheless, small local areas characterized by greater abundance of certain host species and different habitat conditions where ticks are reported to be abundant every season. Practically all our studies were carried on in territory answering the former conditions. The past season was one of only moderate tick infestation.

The season of 1915 was apparently a good year for all species of ticks found in the territory considered.

Economic Conditions Relating to Tick Abundance and Control.

A study of industrial conditions has indicated several points of apparent importance; (1) the principal pursuits are the raising of horses, cattle and sheep, (2) only a small portion of the land is under cultivation, (3) individual holdings of land are relatively large, (4) habitations are mainly found in the valleys near the rivers and creeks. These conditions are likely to be modified to no inconsiderable extent, however, if settlers continue to come into the country as rapidly as they have the past two seasons. Modifications are likely to be apparent along the following lines; (1) an increase in the number of small holdings, (2) an increase in the number of work horses and milk cows, (3) an increase in the number of persons living in regions away from the valleys, (4) an increase in the acreage under cultivation, and (5) a decrease in the amount of range land and the consequent hastening of the time when a great proportion of what is now range stock will be kept within fenced areas.

At present the proportion of horses used for domestic purposes and pastured in proximity to habitations is relatively small. Most horses are those raised for speculation and are pastured on the open range. A few milk cows are gen-

erally kept at the ranches and homesteads, the remainder are range stock. Most cattle men find it necessary to winter their range stock in fenced areas in or close to the valleys. Such cattle are usually in the valley during the first part of the tick season in the spring. They may later be brought in from the range for branding. Horses also may be brought in several times during the tick season. Sheep are frequently driven for considerable distances in the spring from the winter to the summer ranges, often crossing the valleys near habitations. Lambing usually occurs in May, shearing in June. The shearing pens are generally some distance from the ranches.

Crops are grown almost entirely for local consumption. They are mainly corn, alfalfa, and various small grains. Small garden plots are the rule. Hay is cut in considerable quantities, but almost entirely from uncultivated land.

Wild Mammals as Hosts of the Spotted Fever Tick.

A survey of the wild mammalian fauna and its relation to the life history and abundance of the tick was an important feature of the investigations. The results of the season's work, though incomplete, contain considerable information of value. Twenty-six species of wild mammals were found to be present. One thousand and thirty-seven specimens belonging to 22 species were examined for ticks. One thousand and five specimens belong to 11 species that were found to be tick hosts. The names of these animals, the number of each species examined, the number and per cent of each species infested and the stages of the ticks which were found to feed on each are indicated in the accompanying table. The writers are indebted to Mr. E. A. Preble of the United States Biological Survey and to Professor Spaulding of the State College for the identification of most of the mammals.

A comparison of the wild mammals found to be tick hosts in the Bitter Root Valley shows but three species common to both localities, the cottontail rabbit, the coyote and the badger. A still more significant difference, however, is the fact that in eastern Montana we find among the small mammals, an apparently important host of adult ticks. This is the jack rabbit. Of 84 jack rabbits examined 56 or 66.66 per cent were infested with adults and of 68 examined before July 1, 49 or 72.08 per cent. A fully engorged female was

TABLE OF WILD ANIMAL HOSTS OF SPOTTED FEVER TIC IN EASTERN MONTANA.

COMMON NAME	SCIENTIFIC NAME	No. Examined	No. Infested	Per Cent Infested	Larvae	Host of Nymph	Adult
Prairie dog.....	<i>Cynomys ludovicianus</i>	347	29	8.36			
Jack rabbit.....	<i>Lepus townsendi campantus</i>	84	65	77.38	+	+	+
Cottontail rabbit.....	<i>Sylvilagus nutalli grangeri</i>	8	2	25.00	+	+	+
Striped spermophile.....	<i>Citellus tridecemlineatus pallidus</i>	33	10	32.32		+	
Kangaroo rat.....	<i>Peripodops montanus richardsoni</i>	118	24	20.34		+	
Pack rat.....	<i>Neotoma cinerea</i>	7	5	71.43	+	+	
Upland meadow mouse.....	<i>Microtus ochrogaster haydeni</i>	61	6	9.84	+	+	
Grasshopper mouse.....	<i>Onychomys leucogaster missouriensis</i>	19	4	21.05		+	
Deer mouse.....	<i>Peromyscus maniculatus osgoodi</i>	307	37	12.05	+	+	
Pale chipmunk.....	<i>Eutamias pallidus</i>	17	7	41.18	+	+	
Porcupine.....	<i>Erethizon cyprianthus</i>	4	3	75.00	+	+	
American antelope.....	<i>Antilocapra americana</i>	2					+
Pocket mouse.....	<i>Perognathus fasciatus</i>	1					
Pocket gopher.....	<i>Thomomys talpoides bullatus</i>	18					
House mouse.....	<i>Mus musculus</i>	2					
Harvest mouse.....	<i>Reithrodontomys m. dychei</i>	1					
Black footed ferret.....	<i>Mustela nigripes</i>	1					
Badger.....	<i>Taxidea taxus</i>	1					
Skunk.....	<i>Mephitis hudsonica</i>	1					
Coyote.....	<i>Canis l. testes</i>	2					
Bob cat.....	<i>Lynx rufus</i>	2					
Long eared bat.....	<i>Myotis evotis</i>	1					
		<u>1,037</u>					

taken from a jack rabbit as late as August 12. It is perhaps a question what proportion of females become fully engorged on this host, but specimens scarcely more than one-fourth engorged deposited fertile eggs. It is an interesting fact that of 157 adults taken from jack rabbits, 102 were males and only 55 females. Ticks from horses, on the other hand, average about equal numbers male and female. The average number of adult ticks per jack rabbit examined was 1.87, the average per horse 1.44. In one instance 14 adults were taken from a single rabbit. These rabbits are also important as nymphal and perhaps as larval hosts; 36 nymphs have been taken from one rabbit. In another instance, 20 nymphs from one host all molted to adults. Considering the abundance of these animals it is evident that they are an important factor in tick abundance. Porcupines are also hosts of adult ticks, though their importance remains to be determined. In localities where numerous they may be of some interest. A fully engorged female was taken from one of these animals on July 27. The eggs commenced hatching on August 31. Two adults were taken from prairie dogs but their occurrence on this host is considered accidental. Though coyotes examined were found uninfested, it is likely that they are hosts of the adult tick.

Jack rabbits, cottontail rabbits, prairie dogs, chipmunks, deer mice, prairie mice, grasshopper mice, pack rats, and kangaroo rats are hosts of both larvae and nymphs. Spermophiles and porcupines are nymphal hosts. In marked contradistinction to the conditions which exist in the Bitter Root Valley no one species of small mammal, unless it may possibly be the jack rabbit, stands out as of preeminent importance as a nymphal host. Pack rats are efficient hosts but are not present in sufficient numbers to be of great moment. On June 29, 45 larvae and nymphs were taken from one rat. Chipmunks may be important in certain localities where conditions favor their existence in relatively large numbers. Kangaroo rats and deer mice are interesting possibilities, the latter because of their great numbers. Under suitable conditions large numbers of them are found in comparatively small areas. Grasshopper mice are scarce and relatively unimportant. Prairie dogs are abundant locally, mainly on the bench land, and are much less common in the hills. This season's work would indicate that they are not of great importance.

In the paragraph preceeding, several animals have been mentioned as larval hosts. Under laboratory conditions the prairie dog is an extremely efficient larval host. Only one was found infested with larvae in nature during 1916, but larvae were secured from them in 1915, when ticks were more numerous. Cottontails may be important. One hundred and eleven seed ticks were taken from a cottontail on July 31, 1915. Unfortunately only 22 were determined, but all these were *Dermacentor venustus*. During 1916 the great majority of seed ticks were taken from animals of nocturnal habits.

It is of interest to note in connection with the importance of rabbits as adult hosts, that they were very abundant for several years previous to 1915. In 1914 they began to die off and decreased greatly in numbers in 1915. Local residents reported grubs in the backs of thin and sickly-looking rabbits. This probably indicated bot flies, though whether they were responsible for the great mortality is another question. The only rodent bot fly captured in the locality was *Cuterebra tenebrosa* Coquillett. Similar epidemics of a periodic nature have been reported from many parts of the west and northwest.

Another point worthy of mention is the fact that those animals which were found both in rocky situations in the hills and under valley conditions showed the highest percentage of infestation with the immature stages of the tick in the former places. This was particularly noticeable among the deer mice, which showed much lighter infestation under valley conditions. Rock strewn hillsides and rock crowned hills are common among the hills bordering the Powder River Valley.

Certain of the observations made seem to indicate that ticks may sometimes be abundant in the absence of domestic animals. This condition requires further investigation.

Domestic Animals as Tick Hosts.

Under this heading are included horses, cattle, sheep, dogs, cats and pigs. Only adult ticks are known to occur on these animals. One hundred and sixty-one horses were examined and 252 ticks secured, 115 males and 117 females. an average of 1.44 ticks per horse. From fifteen range horses examined in June an average of 6 adults per animal were collected and from 146 work and saddle horses ex-

amined from March 27 to August 27, an average slightly less than one tick per animal. This seems difficult to understand if it indicates the true proportion of ticks that feed on these two types of horses and necessitates a study of condition on the open range to determine what small mammals, under range conditions, engorge the immature stages of the tick. Rabbits may be responsible. One resident reported removing 37 ticks from a single range horse early in July. From a work horse examined 7 times between April 15 and May 18, 33 ticks were secured; other horses in the same pasture had very few ticks on them. In 1915 ticks must have been very abundant on horses. One person described them as so numerous on the bellies of his work horses that they were scraped off repeatedly with a stick. Hand-picking was practiced by many ranchers.

As all cattle except a few milk cows were range stock, opportunities to examine them for ticks were not common. Calves tied or held for branding or vaccination were examined on several occasion. From 150 calves examined April 11, 7 ticks were removed, from 4 on April 17, 1 tick, from 136 on June 7, 5 ticks. In each instance all the ticks taken were females. They were found attached to the brisket behind the forelegs. Those taken on June 7 were kept to see if they would deposit eggs. Though all were more than half engorged but one lot of eggs was deposited and these few in number and not fertile. All the females became abnormal in appearance and soon died. Residents perfectly familiar with the tick and its occurrence on horses, reported never having seen it on cattle. Milk cows were examined several times with negative results. It is difficult to interpret this fact in view of the importance attached to cattle as tick hosts in western Montana and definite conclusions cannot be based on such meagre and unexpected results.

Between April 8 and June 15, 7 sheep were examined. From these 7 adults were secured, 4 males and 3 females. One female was fully engorged, 2 males and 1 female attached, 2 males unattached and 1 male dead in the wool. Several herds sheared during the middle of June were examined by the shearers and in part by us. Ticks were found on but 2 sheep and were unfortunately lost before they were turned over to us for determination. Some of the residents claim ticks to be most abundant in territory

ranged over by sheep, others the opposite. Horses in sheep territory were apparently as badly infested as elsewhere. Sheep owners reported ticks very common on sheep in 1915. It is impossible to make any suggestions concerning the importance of sheep as adult hosts.

A single tick was removed from dogs, though many were examined. The infested dog was herding sheep. Ticks were said to be common on dogs in 1915. Cats and pigs were examined with negative results.

Ticks on Human Beings.

Ticks were common on persons during the season of 1915, considerable numbers sometimes being picked up during one day. Though ticks were relatively far less abundant during 1916, the great majority of persons had found them on their body or clothing at one time or another during the season. Their occurrence on persons may be from early March to at least well toward the middle of August. One of the cases of spotted fever in 1915 occurred in late July, the tick, well engorged, having been removed on July 31. *Dermacentor variabilis* has previously been mentioned as occurring on a man at Kirby, Big Horn County.

Sores caused by ticks were frequently heard of, the bite in several cases having been received in 1915, local irritation being still present. In the cases examined the scars greatly resembled those of small pox vaccination. One case was reported of a boy bitten several years ago, who has quite severe, recurring, local symptoms each spring.

The residents have fully recognized the importance of the tick as the transmitting agent of Rocky Mountain spotted fever and were found to exercise considerable care against ticks becoming attached to their persons.

Life History Notes.

Information concerning the life history of the tick under eastern Montana conditions was gleaned from several sources; engorged females, nymphs and larvae taken from wild and domestic animals, records of engorgements secured from rearing experiments and the subsequent moltings and records of the seasonal occurrence of larvae, nymphs and adults on various host animals. The first larvae were taken on May 18, the last on August 21; the first nymphs on April 10, the last on August 30; the first adults on March 27, the last on August 31. Adults were reported from about

the first of March. The preoviposition period was found to vary from 6 to 14 days (25 records between April 8 and August 21), the oviposition period from 15 to 40 days (23 records between April 17 and August 23), the incubation period from 24 to 51 days (17 records between April 17 and August 31), the period of larval engorgement from $1\frac{3}{4}$ days to $5\frac{1}{2}$ days (4830 records between June 22 and August 21), the larval premolting period from 8 to 26 days (2806 records between May 26 and August 29), the period of nymphal engorgement from 4 to 10 days (194 records between July 3 and August 4), the nymphal premolting period from 15 to 53 days (195 records between May 8 and September 3.) Under experimental conditions it was found impossible to successfully engorge larvae after the middle of August. Nymphs also showed less inclination to attach. Adults were present in greatest numbers from the middle of April to the middle of June. In 1915 they were said to have been abundant till the middle of July.

The tick evidently passes the winter as unfed nymphs and unfed adults, possibly also as engorged nymphs and unfed larvae. This is similar to conditions found in the Bitter Root Valley. Other conditions differ somewhat; the adults appear earlier and stay later, there being no abrupt decline in adult abundance, indicating that a greater percentage of adults engorge after July 1. Wherein the life cycle in the two sections of the State will be found to differ may be expected to hinge to some extent on what happens to the eggs deposited by females engorged during this prolongation of the adult feeding period, and to larvae that may hatch from them. Larval ticks were found about 6 weeks before their normal appearance in the Bitter Root Valley, but whether such earlier occurring larvae had overwintered or hatched from eggs deposited during the spring of 1916 is mere supposition. On the basis of laboratory data the latter possibility is admissible, but it may not be possible under more natural conditions. The period of nymphal premolting (under laboratory conditions) was much shorter than in western Montana and may affect the length of the life cycle in some particulars. Larvae and nymphs show a marked disinclination to feed after the middle of August, adults somewhat earlier.

Longevity Experiments.

In order to increase our knowledge of the life history of the tick and to secure information likely to be of value for control purposes, a series of longevity experiments was started. In nature ticks had been found on animals living under four conditions of flora and soil covering which might have an influence on the longevity of unfed ticks of the several stages. These conditions were as follows; (1) rocky areas, (2) sage brush areas, (3) grassy areas, and (4) areas of small extent occupied by open stands of trees with dense underbrush. Adults and nymphs were released in cages under conditions typifying the first three conditions named and larvae under condition one. Rocks may be considered as affording complete shade, sage brush partial shade and grassy areas no shade. Both cages and longevity tubes were used.

As it was impossible to secure sufficient ticks for these experiments in nature, it was necessary to rear them under laboratory conditions. Thirty-seven cages of animals were infested with seeds and nymphs; 4830 larvae were engorged on prairie dogs, and 1144 nymphs on prairie dogs, jack rabbits, cottontail rabbits, Belgian hares and pack rats. In the actual experimental work 163 adults, 690 engorged nymphs, 1219 unfed nymphs and several hundred larvae were released.

Means by Which Ticks Are Spread.

In eastern Montana there are no apparent natural barriers against the spread of ticks. The limit of spread from any given locality is that of the ranging powers or habits of the host animals. Cattle, horses and sheep are the most important agents of dispersal, due to their unrestricted movements and wide radius of travel. Cattle should perhaps be considered as potential agents until their importance as tick hosts in eastern Montana is more certainly determined. Among small wild animals, jack rabbits are important and are extensive travelers, though their ultimate range is scarcely likely to be as great as that of horses and cattle. Coyotes may also be important.

Due to the untrammelled movements of so many hosts of the adult ticks we would naturally expect that infestation in any given area would be less severe than in fenced or

naturally limited areas of the same size, in which host animals were confined. Similarly the number of infected ticks in any given locality would be likely to be smaller and the chances of human infection correspondingly less. This is the apparent condition which has existed in eastern Montana as compared with the Bitter Root Valley.

HOUSE FLY CONTROL IN MONTANA.

An investigation of house fly conditions in Montana was begun in 1914 and a summary of the results was presented in the First Biennial Report. During the season of 1915 the work was continued in a different form in Miles City and results of much practical value in house fly control were secured.

The house fly is now well known to be a very important factor in the spread of various diseases. Moreover, flies have been exceedingly abundant in Montana and a study of the particular conditions found in Montana was very necessary. So far as could be done without interfering with the work on the spotted fever tick this work has been taken up by Dr. Parker.

We already have sufficient information to enable us to cooperate with cities and individuals in control work. Numerous newspaper articles have been written and lectures have been given.

A condensed summary of the house fly follows:

The House Fly and the Control of Flies.

R. R. Parker, Ph. D.

In the first biennial report of this board (pages 35 to 50) the results of preliminary investigations conducted at Laurel, and other cities in the Yellowstone Valley, concerning the habits and control of the house fly were briefly summarized. Sufficient data were then collected to permit action in an advisory capacity to cities, towns and institutions and individuals interested in control work.

As far as cities in this State are concerned, the following is a general statement of the facts to be considered in outlining fly control:

1. That the house fly mainly breeds in horse manure.
2. That the house fly also breeds in other animal manures (especially pig manure), refuse of various sorts, and human excrement.
3. That unclean conditions serve to attract flies to any given locality.
4. That adequate measures for the control of the house fly will control many other species that are annoying and

troublesome and only less dangerous because of fewer numbers.

5. That fly control should aim,

(a) to prevent egg-laying in so far as possible.

(b) to prevent flies that may be breeding in manure, garbage or other refuse, from maturing within city or town limits, by proper care pending removal and by removal at sufficiently frequent intervals.

(c) to ultimately so dispose of waste that flies breeding therein will not mature.

(d) to so care for materials which attract flies that they will attract them to the least possible extent.

6. The proper disposal of manure is a most difficult problem to meet. Its use agriculturally is not extensive enough nor does it come at the proper time of the year to assist from the standpoint of control. Therefore, so long as measures for its care within city or town limits, and for its removal, do not injure the manure as a fertilizer nor prevent its use as such, any means recommended for its suitable disposal are amply justified.

7. That one condition very difficult to satisfactorily control is the presence of large corrals in or near cities.

8. Control measures should cause the least possible outlay of money and expenditure of time and place the least possible responsibility on the individual that is commensurate with successful results.

9. That no one system of control can be recommended in detail to cover all conditions; only fundamental principles are of general application.

10. Detailed regulations are not necessarily strict but tend to simplify procedure.

11. Health inspectors are an important accessory to intensive control but only feasible in larger cities.

12. Fly control is purely and simply a question of common decency and cleanliness and its intensive and successful application serves to materially better sanitary conditions.

When the above statement of facts is reduced to a working basis, the following subjects are those of most essential importance for regulatory measures.

- (a) The temporary care of horse manure at the point of production, pending removal.
- (b) The removal of horse manure.
- (c) The ultimate disposal of horse manure.
- (d) The temporary care of garbage and refuse at the point of production, pending removal.
- (e) The removal of garbage.
- (f) The ultimate disposal of garbage.
- (g) The elimination of the insanitary privy.

It is important for any community desirous of initiating fly control to recognize the fact that there are two viewpoints from which control may be regarded, (1) as a measure to reduce insects which are an annoyance and pest, (2) as a means of bettering sanitary conditions. If control is attacked from the second viewpoint it is more intensively applied than if approached from the first, but the results likely to be attained from the first will be greatly enhanced.

During the seasons of 1915 and 1916 the fly work, designed to add to our knowledge of the habits and economic importance of flies and to disseminate the practical and scientific application of the same among the people of the State and interested parties, has been pursued along the following lines:

(1) experiments to determine the radius of dispersion of the house fly, (2) lectures (popular and scientific), (3) recommendations for control measures, (4) publications, (5) exhibits, (6) miscellaneous.

Experiments to Determine the Radius of Dispersion of the House Fly. Several investigations have been conducted during recent years for the purpose of securing information concerning the flight of the house fly and its possible limits. The results, however, have been very indefinite as to the actual limits of flight and the number of flies represented in all these experiments total less than 50,000, the largest number used in any one experiment being 2,500. But when we consider that house fly dispersion means the possible spread of flies breeding out at any given breeding area, large or small, it is at once evident that we are concerned with a period of time equivalent to the average life of the fly and with the number of flies which breed out during this interval. Taking the average length of life as from three to four weeks, it is apparent that the numbers of

flies mentioned above would represent but a small portion of those which might emerge from even a small manure pile and that while the results obtained would be relatively correct yet they can by no means be considered as final. Perhaps the general concensus of opinion resulting from such observation as pertain to dispersion under city conditions can best be summed up by the statement, "That the distance flies may travel to reach dwellings is controlled by circumstances. Almost any reasonable distance may be covered by a fly under compulsion to reach food or shelter. When these are close at hand the insect is not compelled to go far and, consequently does not do so." To show the inadvisability of holding to this opinion it is only necessary in view of the summer's results to indicate why the small numbers of flies used in previous experiments constitute a weak point. From our viewpoint flies are considered as spreading outward from a given center, the breeding place. Naturally flies are more abundant at points in relatively close proximity, because, even though they might migrate from this zone their numbers are maintained by the continual emergence of adults. But as they spread outward they must, of necessity, become constantly more and more scattered and their capture increasingly difficult until a point is finally reached the chances of their capture becomes reduced to infinity. But this point is extended further and further with each increase of the number of flies emerging during a given period, the length of which is limited to the average length of life. Consequently we are justified in assuming that experiments dealing with comparatively small numbers are not indicative of finality. With these points in mind and realizing that, for the most part, the conditions in all Montana towns and cities are essentially similar though perhaps differing considerably from those concerned in the experiments mentioned above, the headquarters for the season's observations were located at Miles City which afforded the combination of conditions best suited to the work, namely, abundance of flies, lack of intensively applied control measures and representative size.

It is only possible to give a comparatively brief summary of the work in this paper. A total of 387,877 marked flies were released from four release stations during a period of 35 days. Two of these release stations were situated on

the west side of the city (the sales yards and the city dump), one in the center of the city and one on the east side. Of the marked flies, 1,056 were recaptured at 78 recapture stations, so located that every part of the city was under observation. Marked flies from the sales yards were recaptured at 62 stations out of a possible 69. This figure was higher than that for other release stations due to the fact that a much larger number, 248,140 flies, were released from the sales yards. However, the figures and localities for all release points were such as to show that flies from each one were distributed to very part of the city. Also, flies were not only captured within Miles City, but it is significant that marked flies from all release points were recaptured at the State Industrial School, which is seven hundred yards beyond the eastern limits of the city and the farthest point at which recaptures were attempted. Flies from the city dump and sales yards not only crossed the city but also the 700 yards of open country intervening, in order to reach this point, a total of 3,500 yards (nearly two miles) and 3,070 yards respectively. The longest radius at which flies were recaptured within the city was 2,333 yards (about one and one-third miles) which was the most distant station within the city from any release point.

The investigations, however, were not confined to numbers and locality alone, but also concerned the factors which influence dispersion, that is, those factors which control the direction of flight and the relative abundance in different localities. Wind, temperature, state of weather, etc., are among the factors which have been previously suggested and while under certain conditions and circumstances they unquestionably play their part, they assume less importance when dispersion is considered for long periods of time, unless some special locality offers unusual climatological conditions. A much more practical viewpoint is obtained when we consider the movements of flies to be determined by their reactions to their surroundings; that is, by external stimuli. A discussion of this point is necessarily highly involved and concerns stimuli which cause movements, those which cause inactivity and the conditions under which either kind is dominant. It is sufficient for this report to indicate that movements and consequent dispersion are, in the main, dependent on the stimuli (odors) received from feeding areas

and breeding areas, a statement which is substantiated by the summer's results.

The results above stated indicate the following points of practical significance under city conditions in Montana: (1) That flies from a given breeding area may spread over a territory within a city of at least five square miles, (this is based on the longest radius found within city limits and within which it was found that flies were distributed to all parts), (2) that the actual limit of dispersion within cities may be considerably greater than this, (the number of flies recaptured at the State Industrial School indicates that flies may be abundant even at distances of nearly two miles from their breeding place; that is, that they may spread over a territory of twelve square miles), (3) that flies by no means remain close to their breeding grounds even when food and "shelter" are abundant, but that they lead an extremely migratory existence and will not only cross a city or considerable portions thereof, but will leave it and fly across open country to points some distance beyond; (4) that conditions within a city which are favorable to fly breeding are of importance not only to the residents, but to farmers and others in its vicinity; (5) that even in a city of considerable size, every person who permits conditions favorable to fly breeding to exist on his premises is maintaining a nuisance which is of actual or potential concern to every other person within city limits (this is not only because of the possible migration of flies to any given locality, but also because flies are naturally attracted to stores, dairies and other sources of food supply where they may contaminate food); (6) that the importance of general cooperation in order to secure successful results in control work is strongly emphasized (the cleaning up of a few places here and there has little value); (7) that fly control measures, intensively applied, are far reaching in their effectiveness as a means to help secure general sanitary conditions, both from educational and practical viewpoints; (8) that it is highly necessary to control these conditions which make any locality (feeding or breeding areas) attractive to flies (as indicated in the "First Biennial Report" of the Board the three biggest problems of fly control in Montana are the proper care and disposal of garbage, the proper care and disposal of manure and the elimination of the open privy.)

Lectures. Lectures, (some illustrated), planned to stimulate an interest in fly control, the reasons why it should be undertaken and the benefits to be derived therefrom, have been delivered in the following cities and to the following organizations: Bozeman—Chamber of Commerce, Women's Civic Club, students of the State College, public school teachers, Young Men's Christian Association, and Rebecca's; Havre—Directors of Chamber of Commerce, Chamber of Commerce, Women's "Baby Week" meeting, and public school teachers; Miles City—Chamber of Commerce and Tri-County Teachers' Institute of 1915. Lecture work in both Bozeman and Havre was supplemented by the use of slides in the moving picture houses.

Lectures of a strictly scientific character were delivered before the Science Club of the State College at Bozeman and the Bozeman Medical Association.

In July, 1916, a paper concerning garbage disposal in small towns was read before the State Health Officers Association at Miles City.

Recommendations for Control Measures. Recommendations for fly control have been made to interested organizations in the cities of Bozeman and Havre. These recommendations have been made with the idea that intensive and, therefore, thoroughly efficient control is the most desirable aim, because it must be recognized that measures adopted for fly control are fundamentally measures for good sanitation and for improving conditions detrimental to public health. Hence the nearer it is possible to approach the ideal the most effective will the efforts have been. The carrying out of such measures, as adapted to cities of the size represented, involves an initial financial outlay and the adoption of regulatory measures which of necessity call for adequate time to make the preliminary moves. In both cities noted above there is an earnest disposition to inaugurate active work at the earliest possible moment. Indeed the lectures given in these cities and the active interest of certain local organizations has already done much to improve general sanitary conditions, the value of which will be greatly enhanced under proper direction and supervision.

Suggestions for interesting the townspeople in fly control have been made to interested parties in the town of Hysham.

Control measures recommended to the Montana Power Company are to be adopted at their construction camp at Holter.

The installation of incinerating plants has been recommended when possible. Incineration is unquestionably the most sanitary method for the disposition of garbage.

Requests for advice concerning fly control have also been received from two State institutions, the State Industrial School at Miles City and the State Insane Asylum at Warm Springs. Measures recommended to the former institution were adopted during the summer of 1915 and afforded immediate beneficial results. A visit to Warm Springs was made in June of 1916 and a detailed plan of control submitted.

Publications. Publications concerning flies and fly control may be divided into three groups, (1) newspaper articles, (2) contributions to the Bulletin of the State Board of Health, and (3) scientific papers. A considerable number of newspaper articles have been written, especially as supplementary to lecture work. Under the general heading of "The House Fly in Relation to Public Health in Montana," three articles have been published in the Bulletin of the State Board of Health. The first of these concerned the habits of the house fly and its relation to man, the second discussed its importance as an agent in the distribution of disease organisms and the third dealt with the means of control. A fourth article presented in a popular way the results obtained in the experimental work on dispersion carried on at Miles City.

One article dealing with the dispersion work just mentioned was published in the Journal of Economic Entomology for June, 1916. This paper was purely scientific.

Exhibits. Exhibits illustrating the habits of the house fly and its near relatives were prepared for the State Fair at Helena in 1915 and 1916. These exhibits attracted considerable attention and intelligent interest, attesting the general realization of the importance of such matters in relation to public health.

Miscellaneous. Plans for a maggot trap and for a manure box have been prepared, which may be obtained by application to Professor R. A. Cooley, Secretary of the State Board of Entomology. The maggot trap is an arrangement particularly adapted to country conditions and perhaps af-

fords the best means yet devised for controlling the house fly with a minimum expenditure of time and care on the part of the individual and is at the same time one which permits the most economic preservation of manure which is to be used for fertilizing purposes. The manure box was designed for use under city conditions, and is adapted for use in narrow alleys. The idea incorporated in its construction is adaptable to diverse conditions; the box is built on to the stable, the manure being put into it from within and removed from an outer door. The manure is never exposed out of doors. Its successful use is dependent upon the prompt placing of manure in the box before flies have deposited their eggs on it in the stable and systematic and periodical removal at sufficiently frequent intervals. Such an arrangement will appeal to those who take pride in keeping their premises neat and clean.

Several opportunities have presented themselves for continuing the observations made at Laurel in 1914 and reported in the First Biennial Report concerning flies breeding in human excrement. Additional species of coprophagous flies have been reared.

Notes concerning the seasonal occurrence of certain excreta frequenting flies have also been made, mainly under ranch conditions.

During October an investigation was made at the construction camp of the Montana Power Company at Holter, upon the request of the Secretary of the State Board of Health, to determine the possible responsibility of flies in connection with an outbreak of typhoid fever. This involved certain bacteriological tests which have not yet been completed.

Need of Further Investigation.

Although the subject of fly control is one that is being attacked energetically in all parts of the country there is considerable to be learned concerning its habits and life history that will be of importance from the standpoint of control. In spite of the fact that investigations on this insect are being actively pursued by the United States Bureau of Entomology and in several states, there is no reason to believe that these will solve our problems in Montana. Indeed in those localities where investigations are being most

actively prosecuted the conditions affecting control are utterly different from those in most sections of this state. The question of how the house fly hibernates in Montana is an important one to solve because of its bearing on efficient control measures. Another problem concerns the manner of properly caring for manure during its temporary storage in cities and towns, especially those of small size. Other problems concern the ultimate disposal of manure and garbage and are largely of an economic nature.

